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ORIGINAL PAPER

Nutrient composition of pill millipede manure of the Western Ghats, India

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Abstract: Nutrient composition of the manure of four pill millipedes (*Arthrosphaera dalyi*, *A. davisoni*, *A. fumosa* and *A. magna*) inhabiting in semi-evergreen forests and plantations of the Western Ghats of Southwest India was evaluated. The quantity and quality of fecal pellets differed between millipedes and their habitats (forest and plantation). Organic carbon content in manure was lower in plantations than in forests, while nitrogen content was elevated in plantations. The carbon to nitrogen (C/N) ratio of manure was lower in plantations compared to forests. The phosphorus content in manure was elevated in plantations in all except for *A. dalyi*. Calcium content of manure was increased in plantations than in forests. The contents of magnesium, potassium and phenolics in manure showed varied results. The mass of fecal pellets was correlated only with volume in forests (r=0.882; p <0.01) and pH in plantations (r=0.616; p <0.05), while the volume of fecal pellets was correlated with nitrogen content in forests (r=0.751; p <0.01) and calcium in plantations (r=−0.619; p <0.05). The conductivity was positively correlated with phosphorus and potassium, while magnesium was negatively correlated in forests as well as plantations. Potassium and magnesium were negatively correlated in forests (r=−0.920; p<0.001) and plantations (r=0.692; p <0.05). Overall, the physicochemical characteristics and nutrient composition of fecal pelets differed between millipedes as well as habitats. The low carbon to nitrogen ratio and the increased nitrogen, phosphorus and calcium content in the manure of millipedes inhabiting in plantations indicates possibilities for successfully employing them for *in situ* composting of forest or plantation residues.

Keywords: *Arthrosphaera*; fecal pellets; forests; manure; organic farming; pill millipedes; plantations; Western Ghats

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Introduction

Millipedes are known to feed the plant litter on the forest floor and return about 60% to 90% of organic matter in the form of fecal pellets (Tajovský 1992). For example, nearly 18% to 28% of the annual litter on the forest floor in Zimbabwe will be transformed into millipede fecal pellets and the quantity of feces amounts to 12% of total litter mass (Dangerfield 1993a). Millipede fecal pellet production will be higher in natural habitats than in laboratory microcosms (Dangerfield 1993b). In semi-evergreen woodlands (Guadeloupe, French West Indies), millipede fecal pellets consist of plant fragments, plant epidermal cells, pollen, earthworm cells, bacteria, fungal hyphae, coal, mineral particles, and amorphous organic matter (Loranger et al. 2003). Field-collected millipede manure (*Alloporus uncinatus*) consists of 52% of soil and 48% of organic matter (Mwabvu 1996a). The increase in water-holding capacity as well as microbial activity in fecal pellets is due to decrease in particle size and the surface area of organic matter (Witkamp and Crossley 1966; McBrayer 1973; Hanlon 1981; McGonigle 1995).

The increase in nitrogen content and decrease in C/N ratio between millipede food and feces was reported by Edwards (1974) and Anderson et al. (1983). Millipede manure is known to store more nutrients in habitats with high rates of litter decomposition (e.g., the Cousine Island of Seychelles) and retard the rapid loss of nutrients by leaching and erosion (Dangerfield 1990; Lawrence and Samways 2003). Mwabvu (1996b) opined that field-accumulated fecal pellets of millipedes are more stable than that of pellets generated by laboratory feeding trials. The large pellets with mineral soil were more friable compared to organically rich small pellets (Dangerfield 1993b). However, in cal-

careous soils, fecal pellets of *Glomeris marginata* (temperate pill millipede) were stable due to presence of high calcium content (Kubikov and Rusek 1976).

Studies on pill millipede manure belonging to the genus *Arthrosphaera* in the tropics are relatively meager. In laboratory feeding trials, fecal pellet production was highest on partially decomposed (conditioned) mixed leaf-litter diet than on single leaf-litter diet (Ashwini and Sridhar 2005). In *Arthrosphaera magna*, the rate of leaf-litter ingestion (561.1–598.2 vs. 1.02–157 mg·animal^{-1}·day^{-1}) as well as fecal pellet production (550–570 mg vs. 0.97–126.05 mg·animal^{-1}·day^{-1}) were higher than other millipedes (Ashwini and Sridhar 2005). Similarly, the mass of fecal pellet production by *A. magna* was high in millipedes fed with humus compared to those fed with leaf litter + soil and soil (Ashwini and Sridhar 2006a). Although pill millipedes are found in forests of the Western Ghats, they invade organically managed plantations (Ashwini and Sridhar 2006b).

The objective of our study was to evaluate the nutrient composition of the manure of four pill millipede species belonging to the genus *Arthrosphaera* endemic to the semi-evergreen forests and plantations of the Western Ghats of India. Mass and volume of fecal pellets and qualities of manures (pH, conductivity, organic carbon, nitrogen, C/N ratio, phosphorus, calcium, magnesium, potassium and phenolics) of pill millipedes in forests and plantations were compared with a view of *in situ* organic farming in plantations.

Materials and methods

Study area and sampling

To collect the manure samples of four species of *Arthrosphaera*, mixed forests and nearby plantations located in different altitudinal gradients (91–1384 m above sea level) of the Western Ghats were chosen (Table 1). Pill millipedes in the litter strata of forests and plantations during southwest post-monsoon season (October-November, 2005) were sampled. Five transacts in sampling locations at about 25 m apart were chosen and up to 15 to 20 millipedes from each transact were transferred to perforated polythene boxes (15 × 10 × 10 cm). From there, they were left for about 2 to 3 h without addition of leaf litter, and the accumulated fecal pellets were then separated and shade-dried within 4–6 h of sampling. Once the fecal pellets had attained a constant weight, they were further analysed.

Manure analysis

Randomly selected air-dried fecal pellets were assessed for volume (dial calipers; Mitutoya # 505-626, Japan; accuracy, 0.001 mm) and dry mass (gravimetric; Mettler Toledo, Switzerland; Model PB 303DR). Replicate fecal pellet samples were powdered; mixed with distilled water (1:20 w/v); shaken (15 min.); and pH (Systronics, India, Model 335) followed by electrical conductivity (Systronics, India, Model 304) were determined. Acid-digested manure samples were assessed for organic carbon (Walkley and Black's rapid titration method), total nitrogen (macro-Kjeldahl method), phosphorus (ascorbic acid method) (Jackson 1973), calcium, magnesium, potassium (atomic absorption spectrophotometry: GBC 904AA, Germany) (AOAC 1990) and total phenolics (Folin-Dennis assay) (Rosset et al. 1982).

Data analysis

Differences in the mass, volume, and and chemistry of pill millipede fecal pellets from forest and plantations were assessed by a paired *t*-test (StatSoft 2008). The relationship between mass, volume, and chemistry of pooled data of the four types of millipedes between forests and plantations was assessed based on Pearson correlation (parameters: p values, two tailed; confidence intervals, 95%) (SPSS 16.0: www.spss.com).

Table 1. Forests and plantations of the Western Ghats surveyed for manure of pill millipedes

Location and millipede	Geographical coordinates	Elevation (m, asl)	Vegetation: Forest type	Vegetation: Major plantation crops
Kadaba: *Arthrosphaera dalyi*	12°44′N, 75°29′E	124	Mixed forest (25 tree species)	*Areca catechu, Hopea parviflora, Cinnamomum* sp. and *Piper* sp.
Basrikallu: *Arthrosphaera davisoni*	13°29′N, 75°40′E	1387	Mixed forest (25 tree species)	*Areca catechu*, *Coffea arabica* and *Elettaria cardamomum*
Madikeri: *Arthrosphaera fumosa*	12°25′N, 75°44′E	1147	Mixed forest (21 tree species)	*Areca catechu*, *Cocos nucifera, Coffea arabica, Musa paradisiaca* and *Piper* sp.
Adyanadka: *Arthrosphaera magna*	12°42′N, 75°00′E	91	Mixed forest (15 tree species)	*Areca catechu* and *Cocos nucifera, Musa paradisiaca* and *Theobroma cacao*

Results

Differences in the mass and volume of fecal pellets of *Arthrosphaera* spp. collected from forests and plantations are given in Table 2. Fecal pellet mass (42.8 vs. 24.7 mg; p <0.001) as well as volume (77.2 vs. 36.3 mm^3; p <0.05) of *A. fumosa* was significantly higher in forests than in plantations (Table 1). Fecal pellet mass was least in *A. magna* sampled from forests (16.1 vs. 26.6–42.8 mg) as well as plantations (15.3 vs. 18–32.1 mg). The volume of pellets of *A. dalyi* was lowest in forests (34.2 vs.

38.4–77.2 mm^3), while in *A. fumosa* it was lowest in plantations (36.3 vs. 39.3–48.1 mm^3). Overall, fecal pellet mass (p <0.01) and volume (p <0.001) of *Arthrosphaera* spp. were significantly higher in forests compared to plantations.

Table 2. Dry mass and volume of fecal pellets of pill millipedes in forests and plantations of the Western Ghats

Items	Forest	Plantation
Dry mass per pellet (mg) (n = 25, mean ± SD)		
Arthrosphaera dalyi	26.64±3.81^a	32.11±3.25^b*
A. davisoni	36.28±3.81^a	17.98±2.63^b**
A. fumosa	42.79±3.48^a	24.72±1.36^b***
A. magna	16.06±2.87^a	15.34±1.50^a
Mean (n = 100±SD)	27.92±3.62^a	22.51±2.78^b**
Volume per pellet (mm^3) (n = 25, mean ± SD)		
Arthrosphaera dalyi	34.21±1.58^a	48.11±6.08^b*
A. davisoni	68.63±7.13^a	39.28±2.69^b**
A. fumosa	77.22±6.82^a	36.32±2.15^b*
A. magna	38.41±2.89^a	45.00±2.11^b*
Mean (n = 100±SD)	54.60±4.11^a	42.19±4.66^b***

Notes: Different letters across the columns of each species between forest and plantation differ significantly (*, p<0.05; **, p<0.01; ***, p<0.001).

Variations in chemical composition of manure sampled from forests and plantations are presented in Fig. 1–3. The pH of manure of *A. dalyi* (p <0.01) and *A. fumosa* (p <0.05) was significantly higher in plantations than in forests, while it was the reverse in *A. davisoni* and *A. magna* (p <0.05) (Fig. 1). The pH was close to neutral in manure of *A. davisoni* and *A. magna* sampled from plantations. Conductivity of manure of *A. dalyi* (p<0.05) and *A. davisoni* (p <0.001) was significantly higher in forests than in plantations, while in *A. fumosa* (p<0.01) it was reverse. In *A. magna* (p >0.05) there was no difference in conductivity between forest and plantation.

The organic carbon content of manure of *A. dalyi, A. davisoni* (p<0.05) and *A. fumosa* (p <0.01) was significantly elevated in forests compared to plantations, while *A. magna* possess the lowest organic carbon in both forests and plantations (p >0.05) compared to other millipedes (Fig. 2). Nitrogen content of *A. dalyi*, *A. davisoni* (p <0.05) and *A. fumosa* (p <0.01) manure was significantly higher in plantations than in forests, while there was no significant difference in *A. magna* (p>0.05). Unlike *A. magna* (p >0.05), the C/N ratio was significantly higher in *A. dalyi* (p <0.05)*, A. davisoni* (p <0.01) and *A. fumosa* (p <0.05) in forests than in plantations.

Phosphorous content in *A. dalyi* manure was significantly higher in forests than in plantations (p<0.01), whereas it was the reverse in *A. davisoni*, *A. fumosa* (p<0.05) and *A. magna* (p >0.05). Calcium content in *A. dalyi*, *A. davisoni* and *A. fumosa* manure was significantly elevated in plantations compared to forests (p <0.001) unlike in *A. magna* (p>0.05) (Fig. 3). Magnesium content of *A. dalyi* and *A. davisoni* was significantly higher in plantations than in forests (p <0.01), while it was the opposite in *A. fumosa* (p<0.01) and *A. magna* (p >0.05). Potassium content in *A. dalyi* (p <0.01) and *A. davisoni* (p <0.001) was significantly higher in forests than in plantations, while it was reverse in *A. fumosa* (p <0.01) and *A. magna* (p <0.05). Total phenolics in *A. dalyi* and *A. magna* manure was significantly lower in plantations compared to forests (p <0.01) unlike *A. davisoni* and *A. fumosa* (p <0.001). Overall, the nutrient content of fecal pellets differed between millipedes as well as the habitats studied.

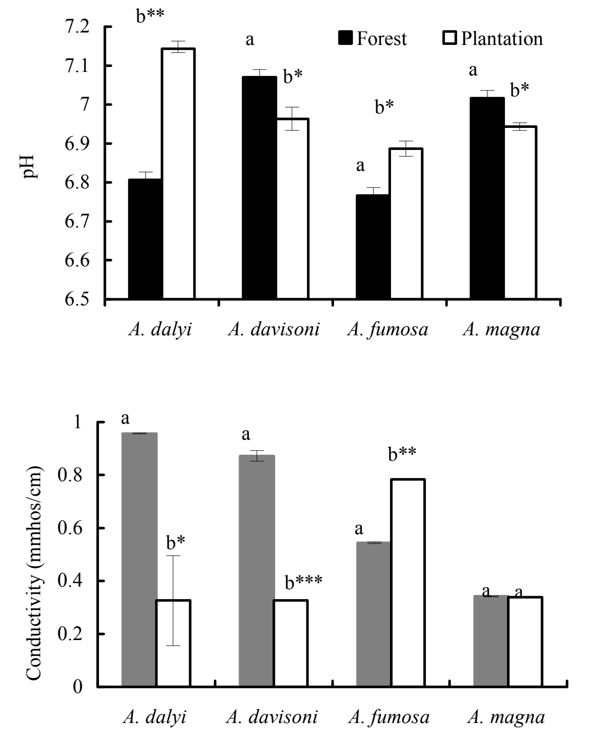

Fig. 1 pH and conductivity of manure of *Arthrosphaera* species in forests and plantations (different letters across the bars indicate significant difference: *, p<0.05; **, p<0.01; ***, p<0.001).

The Pearson correlation between fecal pellet mass and the volume versus chemistry of the combined data of four millipedes in forests and plantations is given in Table 3. Among the nine parameters tested, conductivity of manure correlated most with organic carbon in the forest and the phosphorus, calcium, magnesium and potassium in forests as well as plantations). Fecal pellet mass was positively correlated only with volume in forests (r=0.882; p <0.01) and pH in plantations (r=0.616; p <0.05), while the volume was positively correlated with nitrogen content in forests (r=0.751; p <0.01) and negatively correlated with calcium in plantations (r=–0.619; p <0.05). The pH was negatively correlated with nitrogen in plantations (r=–0.622; p <0.05) and positively correlated with phenolics in forests (r=0.972; p <0.001), while negatively correlated with calcium (forest, r=–0.734; p <0.01; plantation, r=–0.704, p <0.05) and positively correlated with magnesium (forest, r=0.619, p <0.05; plantation, r=0.697, p <0.05) in forests as well as in plantations. Potassium was negatively correlated with magnesium in forests (r=–0.920, p <0.001) as well as in plantations (r=–0.692, p<0.05).

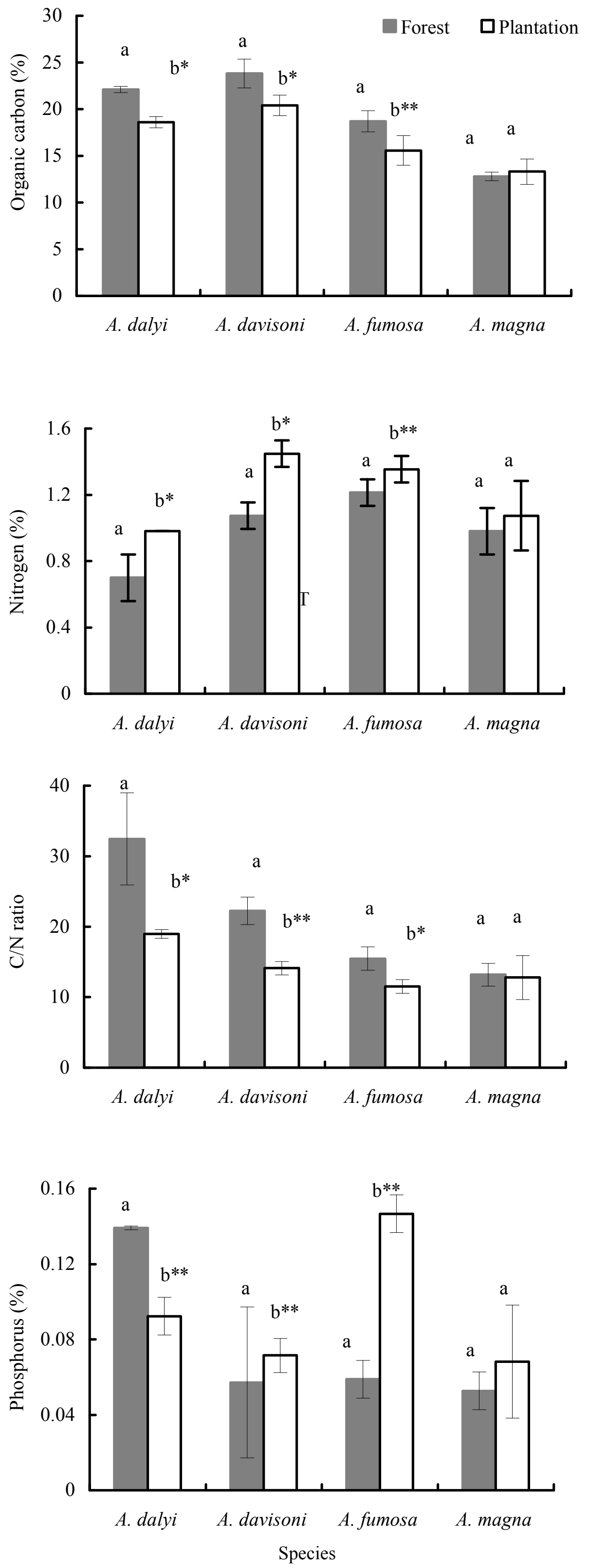

Fig. 2 Organic carbon, nitrogen, C/N ratio and phosphorus of manure of *Arthrosphaera* species in forests and plantations (different letters across the bars indicate significant difference: *, *p*<0.05; **, *p*<0.01).

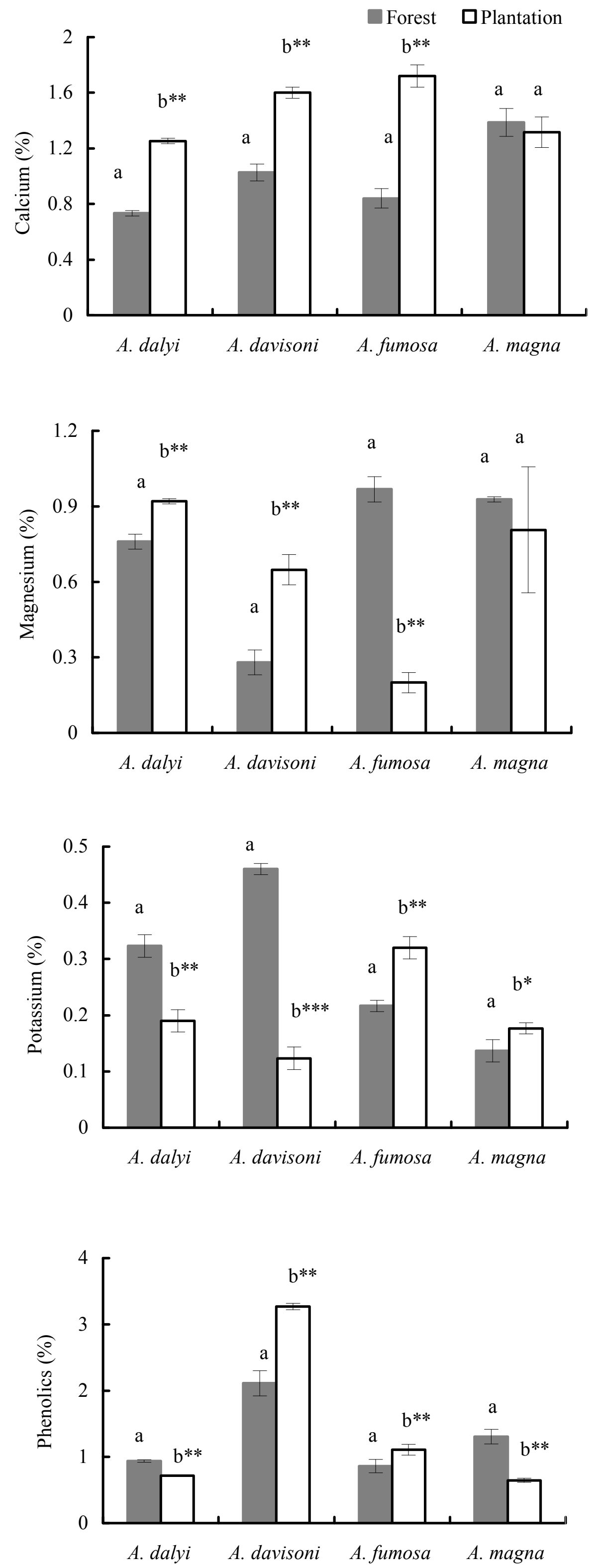

Fig. 3 Calcium, magnesium, potassium and phenolics of manure of *Arthrosphaera* species in forests and plantations (different letters across the bars indicate significant difference: *, *p*<0.05; **, *p*<0.01; *, *p*<0.001).**

Table 3. Pearson correlation coefficients between fecal pellet mass and volume vs. nutrient qualities of pill millipede manure (for combined data of *Arthrosphaera dalyi*, *A. davisoni*, *A. fumosa* and *A. magna*) in forests and plantations (in parenthesis) of Western Ghats (significant correlations are given in bold face)

Index	Mass	Volume	pH	Conductivity	Organic carbon	Nitrogen	Phosphorus	Ca^{++}	Mg^{++}	K^{+}
Volume	**0.882***** (0.528)									
pH	-0.301 (**0.616***)	0.002 (0.510)								
Conductivity	0.266 (0.128)	0.001 (-0.331)	-0.111 (-0.537)							
Organic carbon	0.472 (0.225)	0.308 (-0.097)	-0.086 (0.411)	**0.922***** (-0.288)						
nitrogen	0.473 (-0.354)	**0.751**** (-0.436)	0.061 (**-0.622***)	-0.436 (0.343)	-0.135 (0.297)					
Phosphorus	-0.194 (0.372)	-0.569 (-0.204)	-0.510 (-0.310)	**0.668*** (**0.832****)	0.411 (-0.162)	**-0.769**** (0.359)				
Ca^{++}	-0.534 (-0.185)	-0.207 (**-0.619***)	**0.734**** (**-0.704***)	**-0.703*** (**0.643***)	**-0.702*** (0.213)	0.132 (**0.756****)	**-0.650*** (0.514)			
Mg^{++}	-0.204 (0.060)	-0.282 (0.554)	**-0.619*** (**0.697***)	**-0.649*** (**-0.816****)	**-0.725**** (0.142)	0.031 (-0.458)	0.047 (**-0.592***)	0.057 (**-0.829****)		
K^{+}	0.374 (0.237)	0.314 (-0.347)	0.276 (-0.371)	**0.859***** (**0.842****)	**0.916***** (-0.423)	-0.070 (0.080)	0.212 (**0.872*****)	-0.429 (0.439)	**-0.920***** (**-0.692***)	
Phenolics	0.083 (-0.302)	0.306 (-0.301)	**0.872***** (-0.210)	0.227 (-0.178)	0.297 (**0.670***)	0.153 (**0.715****)	-0.428 (-0.252)	0.384 (0.492)	**-0.853***** (-0.132)	**0.632*** (-0.487)

Notes: Coefficients with asterisk differ significantly (*, p<0.05; **, p<0.01; ***, p<0.001).

Discussion

Pill millipedes belonging to the genus *Arthrosphaera* are fairly common in the Western Ghat forests (Attems 1936; Ashwni and Sridhar 2008; Kadamannaya et al. 2010). Their migration from forests to plantations is evident in different parts of the Western Ghats (Aswhini and Sridhar 2006b; Kadamannaya et al. 2009). They are as efficient as earthworms in feeding on decaying matter, especially in organically managed plantations compared to those managed with chemical fertilizers and pesticides(Ashwini and Sridhar 2002; 2006a; Kadamannaya et al. 2010). Even though the manure of pill millipedes, produced in microcosms, was assessed for its chemical composition (Aswini and Sridhar 2005, 2006a, Kadamannaya and Sridhar 2009), studies are lacking on the manure produced in the forests and plantations of the Western Ghats.

In the giant millipedes of the Seychelles and Southern African forests, the quantity of fecal pellets represented 2.9% and 8 to 13% of the total standing litter, respectively (Dangerfield and Milner 1996; Lawrence and Samways 2003). Interestingly, the rate of mixed litter ingestion by *Arthrosphaera magna* in microcosms was higher than those of temperate millipedes and non-pill millipedes in tropics (561–598 vs. 1–157 mg·animal^{-1}·day^{-1}), so also the quantity of manure (550-570 vs. 1–126 mg·animal^{-1}·day^{-1}) (Ashwini and Sridhar 2005). In laboratory feeding trials, leaf litter ingestion, food conversion efficiency and fecal pellet production by *Arthsosphaera dalyi*, *A. davisoni* and *A. magna* were higher in mixed litter diet than in single litter diet (Ashwini and Sridhar 2005; Kadamannaya and Sridhar 2009). Consumption of humus by *A. magna* yielded the higher amount of manure (670 mg·animal^{-1}·day^{-1}) compared to feeding leaf litter with soil (350–470 mg animal^{-1} day^{-1}) and feeding soil free from leaf litter (30 mg·animal^{-1}· day^{-1}) (Ashwini and Sridhar 2006a).

Millipedes in forests may have wide choice of leaf litter. Usually the leaf fall takes place in the Western Ghats during the winter (December-February), dry up during the summer (March-May) and are conditioned subsequently by microbes during the rainy season (June-November). Such conditioned leaf litter is accessible for feeding by saprophagous fauna and in turn the soil will be enriched. Pill millipedes come out from hibernation shortly after the start of the monsoon (June-July) and are feed on a variety of leaf litter (Ashwini and Sridhar 2006b).

The enriched diet of pill millipedes influences the quantity as well as quality of manure. Pill millipede biomass showed positive correlation with soil organic carbon, pH and minerals (P, K and Ca) in Western Ghat plantations (Ashwini and Sridhar 2008; Kadamannaya et al. 2010). Similarly, food consumption and food conversion efficiencies were positively correlated with the litter nitrogen in *Arthrosphaera dalyi* (Kadamannaya and Sridhar 2009). In spite of acidic soil pH in Western Ghats (Ashwini and Sridhar 2008; Kadamannaya et al. 2009), the manure of four pill millipedes showed pH close to neutral in forests as well as plantations in our study, which indicates improvement in soil quali-

ties of forest and plantations by saprophagous-behaviour pill millipedes.

Among the four millipedes in our study, manure of *A. davisoni* and *A. magna* showed pH close to neutral than *A. dalyi* and *A. fumosa*. Also, manure conductivity was higher, while organic carbon was lower than the soils of forests and plantations (Kadamannaya et al. 2009). It is likely, the forest and plantation crops, as monocots and dicots, influence the soil organic carbon and nutrient contents. In addition, the minerals in manure strongly influenced conductivity in our study. For example, phosphorus and potassium in the manure were positively correlated, while magnesium was negatively correlated with conductivity. However, organic carbon was similar, phosphorus and calcium contents were high and nitrogen, C/N ratio, magnesium and phenolics were low compared to that of manure of pill millipedes fed with leaf litter in microcosms (Kadamannaya and Sridhar 2009).

Organic matter in manure of *A. magna* fed with leaf litters of acacia, banana, cashew and coconut amended with soil was ranged between 70.1% and 75.8% (Ashwini and Sridhar 2006a). Potassium content in manure in the present study was higher than that of soil in forests and plantations of Western Ghats (Kadamannaya et al. 2009). Manure produced by *A. magna* received fresh leaf litter diet consists of higher calcium than those received conditioned litter diet, while manure produced by those fed with conditioned banana, cashew and mixed litter possess high magnesium (Ashwini and Sridhar 2005). Assimilation of more of calcium than magnesium, especially from mixed litter diet, by *A. dalyi* and *A. davisoni* was evident (Kadamannaya and Sridhar 2009). It has been postulated that high phenolics and tannins in leaf litter are responsible for little attraction of pill millipedes toward fresh leaf litter (Sakwa 1974). The quantity of phenolics was significantly decreased in manure compared to leaf litter ingested (Kadamannaya and Sridhar 2009), indicating the role of microbes in the gut of millipedes.

The Western Ghats of Southwest India comprise of several commercial forests (e.g. *Hevea brasiliensis* and *Tectona grandis*) and plantations (e.g., *Anacardium occidentale, Areca catechu, Cinnamomum malabatrum*, *Citrus lemon*, *C. reticulata*, *Coffea arabica*, *C. robusta, Elettaria cardamomum* and *Theobroma cacao*). Organic farming using mixed litter in plantations has the great advantage in attracting pill millipedes from forests. Research pertaining to the advantages of pill millipede invasion of plantations or employing pill millipede compost in plantations needs further study. It is advantageous to follow *in situ* recycling of plantation crop residues with pill millipedes to provide desired nutrients to plantation crops. If suitable agroclimatic conditions prevail or created by agronomic practices for invasion of pill millipedes, the litter turnover and improvement of soil fertility can be achieved. In the present study, manure of *Arthrosphaera magna* derived from the forests and plantations of Adyanadka showed no significant difference in conductivity, organic carbon, nitrogen, C/N ratio, phosphorus calcium and magnesium levels revealing the prevalence of similar conditions in both regions (e.g. moisture regime and availability of mixed litter). However, there was significant difference only in fecal pellet pH, potassium and phenolics between forests and plantations. Thus, incorporation of mixed litter and maintenance of moisture in plantations will be advantageous to extend the activity of pill millipedes beyond their activity in forests (Ashwini and Sridhar 2006b).

Compost generated by *Arthrosphaera magna* with mixed lignocellulosic wastes consists five times the amount of small particles (<5 mm) than control (Ashwini and Sridhar 2006c). Millipede compost showed a shift of pH toward neutral, along with elevated phosphorus, calcium, magnesium, and potassium and a narrow C/N ratio. Significant elevation of total nitrogen and phosphorus between compost produced with and without *A. magna* indicates the significance of composting lignocellulosic wastes by pill millipedes. The compost produced from plantation residues by *A. magna* compensates the deficiency of nutrients in farmyard manure (N, P and K) (Chowdappa et al. 1999, Ashwini and Sridhar 2006d).

Conclusions

Monitoring the quantity and quality of pill millipede manure in different biomes in relation to edaphic factors will be useful to understand the precise conditions required for pill millipedes to generate adequate manure for crop improvement in plantations. Similarly, addition of mixed litter to the plantation floor or basin and maintenance of adequate moisture, especially during post-monsoon and summer seasons, will attract pill millipedes from forests to enrich the soil for extended periods. Based on wide litter preference, high fecal pellet mass and improved manure qualities (e.g., phosphorus, nitrogen and minerals), suitable species of *Arthrosphaera* having widely adapted to Western Ghats will be advantageous to sustain ecofriendly agroforestry.

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